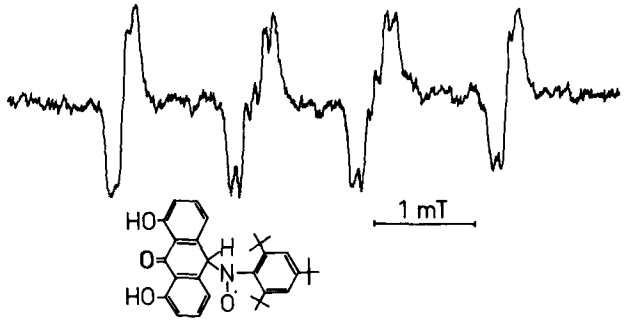
interesting that in spite of its bulky t-butyl groups TBNB acts as a spin trapping agent for such a large radical. It could be expected that the adduct would prefer the sterically less hindered anilino-type structure. This radical may be the active therapeutic form of dithranol. Swanbeck and Thyresson have calculated that dithranol interacts with DNA by intercalation between every 8th base-pair of the double-stranded DNA molecule[7].



The ESR-spectrum of the trapped dithranol radical produced by autoxidation in pyridine. The spectrum was recorded 4 min after dissolution. A Varian E-4 ESR-spectrometer with 100-kHz field modulation was used.

In dilute solutions containing oxygen, the adduct radical is stable enough for ESR-measurement. Within few hours it converts, however, into several different radicals, not trapped by TBNB. The ESR-spectra of these radicals appear in succession at $g = 2.0050$. In more concentrated (~ 3 mmoles/l) dithranol solutions, their formation rate increases and very strong ESR-signals are recorded. The last ESR-spectrum recorded after completion of the oxidation is a very intense singlet, and the corresponding solution has acquired a dark-brown color similar to the 'dithranol brown' well-known to the dermatologists[3]. The work is continued with identification of the latter radicals.

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Influence of treatment duration on audiogenic seizure susceptibility during barbiturate withdrawal in rats

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Summary. Barbiturate withdrawal seizure susceptibility in rats increased with increasing duration of treatment during a 15-day treatment period in which the animals were given an i.p. dose of sodium barbital every 12 h. This method of producing dependence has clear advantages over previously described methods.

In the study of barbiturate dependence phenomena investigators have put forth considerable effort in producing drug dependence in animals, sometimes requiring weeks or months of treatment[1-4]. In these experiments sodium barbital was administered to the animals in their drinking water. Although this method of administration of drug is a relatively simple procedure, it results in some variation in barbiturate blood levels produced[4] since it is impossible to measure and adjust for the amount of drug which is lost by spillage and dribbling or variations in fluid intake by the animals. Since some degree of dependence has been reported after short term treatment of animals with barbiturates[5], the present experiment was conducted to determine the relationship between barbital treatment duration and withdrawal-induced audiogenic seizure susceptibility.

Method. Male rats of Sprague-Dawley descent weighing 200 g (±20 g) at initiation of treatment, and which had been previously screened to be nonresponsive to audiogenic seizure stimulus were used in the study.

Audiogenic response score (ARS) was measured by the method of Jobe et al.[6], in which animals were exposed to high intensity sound (approximately 120 db) produced by 2 electric fire bells inside a testing chamber approximately 40 cm in diameter. In this method the score is determined by visual observation and depends on the number of running fits which characteristically precede the convulsion, and the extent of clonus and/or tonus during the convulsive phase of the seizure. The possible scores range from 1 to 9, with a score of 1 being assigned to a seizure consisting of running fits only, with no convulsive episode. A score of 9 represents a seizure consisting of 1 running fit terminated by a tonic convulsion involving full tonus of the torso and all limbs of the animal.

Test animals received i.p. injections of sodium barbital, 150 mg/kg every 12 h for treatment periods of 5, 10, or 15 days. Control animals received a comparable regimen of normal saline. At the end of the treatment period, injec-

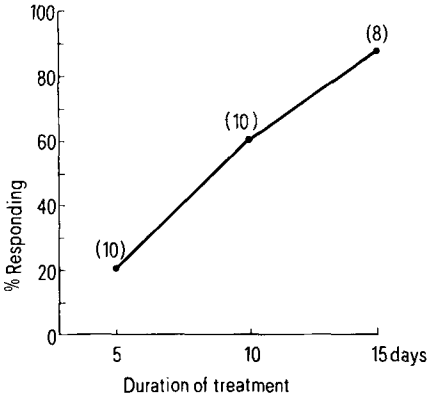

Fig. 1. Relationship between treatment duration and percent of animals responsing (exhibiting any degree of audiogenic seizure susceptibility)during withdrawal. Numbers in parentheses indicate the number of animals tested. Normal saline-treated controls were tested for each time period and found to be unresponsive to the stimulus (see text).

tions were discontinued and ARS determined at 36 h following the last dose.

Results. A linear treatment duration-response relationship was demonstrated for the barbiturate-withdrawn animals, seizure susceptibility (percent responding) increasing with treatment duration (figure 1). No change in seizure severity accompanied the increased susceptibility (figure 2), nor was any spontaneous seizure activity observed. With the exception of 1 rat in the 10-day control group, which had an ARS of 3, control animals exhibited no response to sound stimulus.

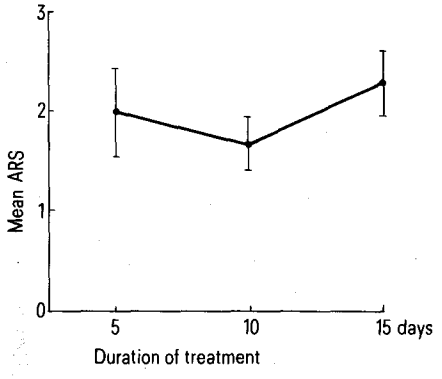

Fig. 2. Effect of barbiturate treatment duration on audiogenic response score (ARS) during withdrawal. Vertical bars represent SE of the mean.

These results provide evidence in agreement with the suggestion of others[5-7] that the physiological changes responsible for barbiturate dependence begin after only a short period of drug treatment. However, even at a sustained high dose level (i.e., the regimen used in this study) a time-dependent process is involved. This partially clarifies the results of a previous report from this laboratory[4], in which it was not possible to state whether the increased seizure response that accompanied progressive weekly increases in barbital dosage was a result of longer treatment or larger daily doses of barbital.

In addition, these results suggest the possibility that the lengthy and variable oral dosing regimens which have been widely employed in the past to produce barbiturate dependence may be an unnecessary complicating factor in investigating the phenomenon.

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Centrally acting hypotensive fraction in the venom of *Dendroaspis angusticeps*

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Summary. I.v. administration of 100 µg/kg of *Dendroaspis angusticeps* venom produced a biphasic vasodepressor response. The first fall in blood pressure is due to a cholinergic component, whereas the second fall may be due to central depressant effect.

In our previous work on *Dendroaspis jamesoni* venom, a central locus of action for the hypotensive response was suuggested[4,5]. This paper deals with the identifaction and mechanism of a hypotensive fraction in *Dendroaspis angusticeps* venom.

Materials and method. 32 cats (2.5-3.5 kg) were anaesthetized with ether followed by i.v. chloralose (80 mg/kg). The blood pressure was recorded from the right common carotid artery by a Statham transducer (P23D) and the heart rate on a Grass polygraph (Model 79-8P-40). The cats were artificially ventilated with an electronic ventilator (SRI, England) at a pressure of 15 cm of water per kg and a rate of 20 per min. The rectal temperature was maintained between 36 and 37 °C throughout the experiment. The efferent pathways of hypotensive response were determined in bilateral cervical vagotomized and spinalized (C-2) cats. The drugs and snake venom were injected through the right femoral vein. Electrophoresis was carried out on starch gel[5] and the protein content of the eluted fractions was determined by the method of Lowry et al.[6].

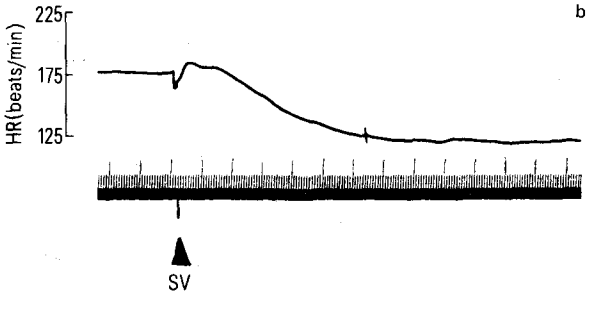

Fig. 1. Effect of i.v. injection of snake venom (100 µg/kg) on the blood pressure in chloralose anaesthetized (80 mg/kg) cat. Cat 2.8 kg, record of blood pressure after i.v. injection of snake venom. Note the initial quick fall, the partial recovery and the sustained fall.